Paper alert

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A selection of interesting papers published in recent months in those major journals most likely to report significant results in protein and RNA folding and design.

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Folding & Design 1996, 1:R89–R94

☐ X-ray structures of a designed binding site in trypsin show metal-dependent geometry. Linda S Brinen, W Scott Willett, Charles S Craik and Robert J Fletterick. *Biochemistry* 35, 5999–6009.

The three-dimensional structures of complexes of trypsin N134H, E151H bound to ecotin A86H are determined at 2.0 Å resolution via X-ray crystallography in the absence and presence of the transition metals Zn^{2+}, Ni^{2+} and Cu^{2+}. The binding site for these transition metals was constructed by substitution of key amino acids with histidine at the trypsin–ecotin interface in the S2′/P2′ pocket. Three histidine sidechains, two on trypsin at positions 143 and 151 and one on ecotin at position 86, anchor the metals and provide extended catalytic recognition for substrates with His in the P2′ pocket. Of the three metals, the binding of zinc results in the most favourable binding geometry, not dissimilar to those observed in naturally occurring zinc-binding proteins.
14 May 1996. *Biochemistry.*

☐ Towards meeting the Paracelsus Challenge: the design, synthesis, and characterization of Paracelsin-43, an α-helical protein with over 50% sequence identity to an all-β protein. David T Jones, Claire M Moody, Julia Uppenbrink, John H Viles, Paul M Doyle, C John Harris, Laurence H Pearl, Peter J Sadler and Janet M Thornton. *Proteins* 24, 502–513.

In response to the Paracelsus Challenge, the authors present the design, synthesis and characterization of a helical protein, whose sequence is 50% identical to that of an all-β protein. The new sequence was derived by applying an inverse protein folding approach, in which the sequence was optimized to 'fit' the new helical structure, but constrained to retain 50% of the original amino acid residues. Although the designed sequence has little ordered secondary structure in water, circular dichroism and nuclear magnetic resonance data show clear evidence for significant helical content in water/ethylene glycol and in water/methanol mixtures at low temperatures, as well as melting behaviour indicative of cooperative folding.
April 1996. *Proteins: Structure, Function and Genetics.*

☐ An engineered allosteric switch in leucine-zipper oligomerization. Lino Gonzalez Jr, Joseph J Plecs and Tom Alber. *Nat. Struct. Biol.* 3, 510–515.

The authors engineered a GCN4 leucine zipper mutant that switches from two to three strands upon binding the hydrophobic ligands cyclohexane and benzene. The crystal structure of the peptide–benzene complex shows a single benzene molecule bound at the engineering site in the core of the trimer. These results indicate that coiled coils are well suited to function as molecular switches and emphasize that core packing is an important determinant of oligomerization specificity.
June 1996. *Nature Structural Biology.*

☐ The single Cys_2–His_2 zinc finger domain of the GAGA protein flanked by basic residues is sufficient for high-affinity specific DNA binding. Paolo V Pedone, Rodolfo Ghirlando, G Marius Clore, Angela M Gronenborn, Gary Felsenfeld and James G Omichinski. *Proc. Natl. Acad. Sci. USA* 93, 2822–2826.

In this paper the authors demonstrated that the minimal domain of GAGA required for specific binding (residues 310–372) includes a single zinc finger of the Cys_2–His_2 family and a stretch of basic amino acids located on the N-terminal end of the zinc finger. It had previously been thought that a single zinc finger of the Cys_2–His_2 family is incapable of specific, high-affinity binding to DNA. The combination of an N-terminal basic region with a single Cys_2–His_2 zinc finger in the GAGA protein can thus be viewed as a novel DNA-binding domain. This raises the possibility that other proteins carrying only one Cys_2–His_2 finger are also capable of high-affinity specific binding to DNA.
April 1996. *Proceedings of the National Academy of Sciences USA*

☐ The designed protein M(II)-Gly-Lys-His-Fos(138–211) specifically cleaves the AP-1 binding site containing DNA. Catherine Harford, Suree Narindrasorasak and Bibudhendra Sarkar. *Biochemistry* 35, 4271–4278.

A new specific DNA cleavage protein, Gly-Lys-His-Fos(138–211), was designed, expressed and characterized. The DNA-binding component of the design uses the basic and leucine zipper regions of the leucine zipper Fos, which are represented by Fos(138–211). The DNA cleavage moiety was provided by the design of the amino-terminal Cu(II)-, Ni(II)-binding site GKH at the amino terminus of Fos(138–211). Binding of Cu(II) or Ni(II) by the protein

activates its cleavage ability. The GKH motif was predicted to form a specific amino-terminal Cu(II)-, Ni(II)-binding motif as previously defined. This prediction was verified as the tripeptide, GKH, and the expressed protein, GKH–Fos(138–211), were both shown to be capable of binding Cu(II) and Ni(II). The designed protein upon heterodimerization with Jun(248–334) was shown to bind to and cleave several forms of DNA which contained an AP-1 binding site. The cleavage was shown to be specific. This design demonstrates the versatility of the amino-terminal Cu(II)-, Ni(II)-binding motif and the variety of motifs which can be generated. The site of cleavage by GKH–Fos(138–211) on DNA provides further information regarding the bending of DNA upon binding to Fos–Jun heterodimers.
9 April 1996. *Biochemistry.*

☐ A 'litmus test' for molecular recognition using artificial membranes. Deborah Charych, Quan Cheng, Anke Reichert, Geoffrey Kuziemko, Mark Stroh, Jon O Nagy, Wayne Spevak and Raymond C Stevens. *Chem. Biol.* 3, 113–120.

Sensitive and selective molecular recognition is important throughout biology. Certain organisms and toxins use specific binding at the cell surface as a first step towards invasion. A new series of biomolecular materials, with novel optical and interfacial properties, have been designed to sense molecular recognition events. Gangliosides that specifically bind cholera toxin, heat-labile *Escherichia coli* enterotoxin and botulinum neurotoxin were incorporated into a matrix of diacetylenic lipids, 5–10% of which were derivatized with sialic acid. The lipids were self-assembled into Langmuir–Blodgett layers and polymerized with UV irradiation, yielding a polydiacetylene membrane with a characteristic blue colour into which the ganglioside is noncovalently incorporated. When toxin is added, the polymerized membrane turns red. The response is specific and selective and can be quantified by visible absorption spectrophotometry. The speed, sensitivity and simplicity of the design offers a new and general approach towards the direct colorimetric detection of a variety of different molecules.
February 1996. *Chemistry & Biology.*

☐ Context-dependent secondary structure formation of a designed sequence. DL Minor and PS Kim. *Nature* 380, 730–734.

The authors have modified the sequence of a region belonging to the second β-hairpin of the B1-domain of the IgG binding protein and they have placed the modified sequence in the α-helix of the same protein. The two proteins, with the modified sequence at the hairpin and at the α-helix, fold into a native conformation. Although this is an interesting result, it is not unexpected since the sequence chosen by the authors does not adopt any particular conformation in solution, and there are many examples in the database of short fragments that adopt different secondary structures in different environments. It seems that non-local interactions can determine the secondary structure of peptide sequences of substantial length.
25 April 1996. *Nature.*

☐ Barriers to protein folding: formation of buried polar interactions is a slow step in acquisition of structure. CD Waldburger, T Jonnson and RT Sauer. *Proc. Natl. Acad. Sci. USA* 93, 2629–2634.

The authors have analyzed the role that the formation of a buried salt bridge plays in the folding reaction of the Arc repressor. The two wild-type buried charge residues have been substituted for hydrophobic ones and the unfolding and refolding kinetics of the wild-type and mutant proteins investigated. The authors have found that the presence of charged residues in the interior of the protein significantly slows down the folding reaction, compared to the mutant with hydrophobic residues. Refolding experiments, performed in the presence of different ionic strengths or viscosities, show that high ionic strength favours folding of the wild-type protein, while high viscosity delays folding of the hydrophobic mutant. The authors suggest that at least for small proteins folding should not be slow and consequently that interactions like the ones mentioned here could be responsible for the slow folding reaction of some of these molecules. It remains to be seen whether slow folding has any functional significance, or if *in vivo* there is an acceptable folding rate window which englobes fast and slow folders.
April 1996. *Proceedings of the National Academy of Sciences USA.*

☐ A new approach to the study of transient protein conformations: the formation of a semiburied salt link in the folding pathway of barnase. M Oliveberg and AR Fersht. *Biochemistry* 35, 6795–6805.

In this work, the authors have studied the changes in the pH dependence of the kinetics of folding of barnase upon mutation of either Asp93 or Arg69, which form a semiburied salt bridge. Opposite to the wild-type protein, the folding behaviour of the Asp93→Asn mutant protein resembles that of a two-state process. The results obtained by the authors lead them to propose that formation of ionic interactions within clusters of hydrophobic residues could control kinetically the folding pathway and play an important role in the early events of protein folding. They also propose that buried salt bridges do not play an energetic role but that it is possible that they add structural specificity to the native conformation. A similar role for buried sidechain hydrogen bonds has been proposed by DeGrado and co-workers. Interestingly enough, in the above selected paper (Waldburger *et al.*, 1996), the authors do not find any structural role for the semiburied salt bridge in the folding of the Arc repressor. It is possible that semiburied or buried salt bridges could play different roles in different proteins, ranging from guiding the early folding events to slowing down folding or determining important functional aspects *in vivo*.
28 May 1996. *Biochemistry.*

☐ Using buried water molecules to explore the energy landscape of proteins. Vladimir P Denisov, Jörg Peters, Hans Dietrich Hörlein and Bertil Halle. *Nat. Struct. Biol.* 3, 505–509.

Buried water molecules constitute a highly conserved, integral part of nearly all known protein structures. Such water molecules exchange with external solvent as a result of protein conformation fluctuations. The authors report here the results of water ^{17}O and ^{2}H magnetic relaxation dispersion measurements on wild-type and mutant bovine pancreatic trypsin inhibitor in aqueous solution at 4–80°C. These data lead to the first determination of the exchange rate of a water molecule buried in a protein. The strong temperature dependence of this rate is ascribed to large-scale conformational fluctuations in an energy landscape with a statistical ruggedness of ~10 kJ mol^{-1}.

6 June 1996. *Nature Structural Biology.*

☐ Use of empirically derived atomic packing preferences to identify favourable interaction regions in the binding sites of proteins. Roman A Laskowski, Janet M Thornton, Christine Humblet and Juswinder Singh. *J. Mol. Biol.* 259, 175–201.

A new empirically based method for predicting favourable interaction regions within the binding sites of proteins is presented. The method uses spatial distributions of atomic contact preferences derived from a non-homologous dataset of 83 high-resolution protein structures. The contact preferences are obtained for 26 different atom types relative to 163 different types of three-atom fragments. Each fragment consists of a triplet of bonded atoms, 1–2–3, which defines a reference frame for the three-dimensional distributions. In this way, directional, as well as distance, information is retained. Once derived, the distributions can be applied in a predictive manner. Given a protein's binding site, each distribution is transformed on to the three-atom fragments of the constituent residues and, when combined, can identify the favourable interaction regions for each different atom type. These predicted regions can then form the basis either for the modification of known inhibitors or for the search and design of new ones. Five known protein–ligand complexes are used to demonstrate the validity and usefulness of the approach. The results show that the method provides a powerful tool both in understanding how a given ligand exploits the interactions available to it in an active site and in helping to design improved, or novel, protein ligands.

31 May 1996. *Journal of Molecular Biology.*

☐ Constructing amino acid residue substitution classes maximally indicative of local protein structure. Michael J Thompson and Richard A Goldstein. *Proteins* 25, 28–37.

Using an information theoretic formalism, the authors optimize classes of amino acid substitution to be maximally indicative of local protein structure. Their statistically derived classes are loosely identifiable with the heuristic constructions found in previously published work. However, while these other methods provide a more rigid idealization of physicochemically constrained residue substitution, their classes provide substantially more structural information with many fewer parameters. Moreover, these substitution classes are consistent with the paradigmatic view of the sequence-to-structure relationship in globular proteins which holds that the three-dimensional architecture is predominantly determined by the arrangement of hydrophobic and polar sidechains with weak constraints on the actual amino acid identities. These substitution classes have been used in highly accurate predictions of residue solvent accessibility. They could also be used in the identification of homologous proteins, the construction and refinement of multiple sequence alignments, and as a means of condensing and codifying the information in multiple sequence alignments for secondary structure prediction and tertiary fold recognition.

May 1996. *Proteins: Structure, Function and Genetics.*

☐ Improving the quality of NMR and crystallographic protein structures by means of a conformational database potential derived from structure databases. John Kuszewski, Angela M Gronenborn and G Marius Clore. *Protein Sci.* 5, 1067–1080.

A new conformational database potential involving dihedral angle relationships in databases of high-resolution highly refined protein crystal structures is presented as a method for improving the quality of structures generated from NMR data. The rationale for this procedure is based on the observation that uncertainties in the description of the nonbonded contacts present a key limiting factor in the attainable accuracy of protein NMR structures and that the nonbonded interaction terms presently used have poor discriminatory power between high- and low-probability local conformations. The idea behind the conformational database potential is to restrict sampling during simulated annealing refinement to conformations that are likely to be energetically possible by effectively limiting the choices of dihedral angles to those that are known to be physically realizable. In this manner, the variability in the structures produced by this method is primarily a function of the experimental restraints, rather than an artifact of a poor nonbonded interaction model. The authors tested this approach with the experimental NMR data. Incorporation of the conformational database potential into the target function used for refinement results in a significant improvement in various quantitative measures of quality (Ramachandran plot, sidechain torsion angles, overall packing). This is achieved without compromising the agreement with the experimental restraints and the deviations from idealized covalent geometry, and the agreement between calculated and observed ^{1}H chemical shifts that provides an independent NMR parameter of accuracy.

June 1996. *Protein Science.*

☐ Energy functions that discriminate X-ray and near-native folds from well-constructed decoys. Britt Park and Michael Levitt. *J. Mol. Biol.* 258, 367–392.

This study generates ensembles of decoy or test structures for eight small proteins with a variety of different folds. Between 35 000 and 200 000 decoys were generated for each protein using a four-state off-lattice model together with a novel relaxation method. These give compact self-avoiding conformations each constrained to have native secondary structure. Ensembles of these decoy conformations were used to test the ability of several types of empirical contact, surface area and distance-dependent energy functions to distinguish between correct and incorrect conformations. These tests have shown that none of the functions is able to distinguish consistently either the X-ray conformation or the near-native conformations from others which are incorrect. Certain combinations of two of these energy functions were able, however, consistently to identify X-ray structures from amongst the decoy conformations. These same combinations are better also at identifying near-native conformations, consistently finding them with a 100-fold higher frequency than chance. The fact that these combination energy functions perform better than generally accepted energy functions suggests their future use in folding simulations and perhaps threading predictions.
3 May 1996. *Journal of Molecular Biology.*

☐ Chaperonin-facilitated protein folding: optimization of rate and yield by an iterative annealing mechanism. Matthew J Todd, George H Lorimer and D Thirumalai. *Proc. Natl. Acad. Sci. USA* 93, 4030–4035.

The authors develop a heuristic model for chaperonin-facilitated protein folding, the iterative annealing mechanism, based on theoretical descriptions of 'rugged' conformational free energy landscapes for protein folding, and on experimental evidence that (i) folding proceeds by a nucleation mechanism whereby correct and incorrect nucleation lead to fast and slow folding kinetics, respectively, and (ii) chaperonins optimize the rate and yield of protein folding by an active ATP-dependent process. The chaperonins GroEL and GroES catalyze the folding of ribulose bisphosphate carboxylase at a rate proportional to the GroEL concentration. Kinetically trapped folding-incompetent conformers of ribulose bisphosphate carboxylase are converted to the native state in a reaction involving multiple rounds of quantized ATP hydrolysis by GroEL. The authors propose that chaperonins optimize protein folding by an iterative annealing mechanism; they repeatedly bind kinetically trapped conformers, randomly disrupt their structure and release them in less folded state allowing substrate proteins multiple opportunities to find pathways leading to the most thermodynamically stable state. By this mechanism, chaperonins greatly expand the range of environmental conditions in which folding to the native state is possible. The authors suggest that the development of this device for optimizing protein folding was an early and significant evolutionary event.
April 1996. *Proceedings of the National Academy of Sciences USA.*

☐ Toward a mechanism for GroEL·GroES chaperone activity: an ATPase-gated and -pulsed folding and annealing cage. Fernando J Corrales and Alan R Fersht. *Proc. Natl. Acad. Sci. USA* 93, 4509–4512.

Free GroEL binds denatured proteins very tightly: it retards the folding of barnase 400-fold and catalyzes unfolding fluctuations in native barnase and its folding intermediate. GroEL undergoes an allosteric transition from its tight-binding T-state to a weaker-binding R-state on the cooperative binding of nucleotides (ATP/ADP) and GroES. The preformed GroEL·GroES·nucleotide complex retards the folding of barnase by only a factor of 4, and the folding rate is much higher that the ATPase activity that releases GroES from the complex. Binding of GroES and nucleotides to a preformed GroEL·denatured-barnase complex forms an intermediately fast-folding complex. The authors propose the following mechanism for the molecular chaperone. Denatured proteins bind to the resting GroEL·GroES·nucleotide complex. Fast-folding proteins are ejected as native structures before ATP hydrolysis. Slow-folding proteins enter chaperoning cycles of annealing and folding after the initial ATP hydrolysis. This step causes transient release of GroES and formation of the GroEL·denatured-protein complexes with higher annealing potential. The intermediately fast-folding complex is formed on subsequent rebinding of GroES. The ATPase activity of GroEL·GroES is thus the gatekeeper that selects for initial entry of slow-folding proteins to the chaperone action and then pumps successive transitions from the faster-folding R-states to the tighter-binding/stronger-annealing T-states. The molecular chaperone acts as a combination of folding cage and an annealing machine.
April 1996. *Proceedings of the National Academy of Sciences USA.*

☐ Effect of GroEL on the re-folding kinetics of α-lactalbumin. Kumiko Katsumata, Akira Okazaki and Kunihiro Kuwajima. *J. Mol. Biol.* 258, 827–838.

The effect of GroEL on the re-folding kinetics of apo- and holo-α-lactalbumin from the acidic molten globule state has been investigated by stopped-flow florescence measurements. GroEL retards the re-folding of apo-α-lactalbumin by interacting with the molten globule state of the protein. The binding constant was estimated to be in the order of 10^5 M^{-1} by analyzing the kinetic data quantitatively and was found to be much weaker than the binding between GroEL and disulfide-bond-reduced α-lactalbumin, whose binding constant is in the order of 10^7 M^{-1}. These results, together with previous results, suggest that the state recognized by GroEL is not unique and that the binding strength varies with the state of a target protein. The binding between GroEL and the molten globule state of apo-α-lactalbumin becomes stronger with an increasing salt concentration; the binding constant is increased 10-fold (from 10^5 to 10^6 M^{-1}) by an increase in salt concentration from 0.05 to 0.25M. The study of the effect of GroEL on the re-folding kinetics of holo-α-lactalbumin, which is represented by a bi-phasic process, shows that the slow

phase is affected by GroEL in the same manner as observed in the apo-α-lactalbumin re-folding but that the fast phase is not affected by GroEL at all. This indicates that the binding rate of GroEL is faster than the slow phase but slower than the fast phase of the re-folding, and the bi-molecular rate constant of GroEL binding to the molten globule state of α-lactalbumin was estimated to be in the order of 10^6 M^{-1} s^{-1}.
24 May 1996. *Journal of Molecular Biology.*

☐ Dynamics of the GroEL–protein complex: effects of nucleotides and folding mutants. Helmut Sparrer, Hauke Lilie and Johannes Buchner. *J. Mol. Biol.* 258, 74–87.
Chaperonins are a ubiquitous class of ring-shaped oligomeric protein complexes that are of crucial importance for protein folding *in vivo*. Analysis of the underlying functional principles had relied mainly on model proteins the (un)folding of which is dominated by irreversible side-reactions. The authors here used maltose-binding protein (MBP) as a substrate protein for GroEL, since the refolding of this protein is completely reversible and thus allows a detailed analysis of the molecular parameters that determine the interaction of GroEL with non-native protein. They show that MBP folding intermediates are effectively trapped by GroEL in a diffusion-controlled reaction. This complex is stabilized via unspecific hydrophobic interactions. Stabilization energies for wild-type MBP increase linearly with ionic strength from 50 kJ mol^{-1} to 60 kJ mol^{-1}. Depending on the intrinsic folding rate and the hydrophobicity of the substrate protein, the interaction of GroEL with MBP folding intermediates leads to a dramatically decreased apparent refolding rate of MBP (wild-type) or a complete suppression of folding (MBP folding mutant Tyr283→Asp). On the basis of this data, a quantitative kinetic model of the GroEL-mediated folding cycle is proposed, which allows simulation of the partial reactions of the binding and release cycles under all conditions tested. In the presence of ATP and non-hydrolyzable analogues, MBP is effectively released from GroEL, since the overall dissociation constant is reduced by three orders of magnitude. Interestingly, binding of nucleotide does not change the off-rate by more than a factor of 3. However, the on-rate is decreased by at least two orders of magnitude. Therefore, the rebinding reaction is prevented and folding occurs in solution.
26 April 1996. *Journal of Molecular Biology.*

☐ Structure of the C-terminal end of the nascent peptide influences translation termination. Asgeir Björnsson, Salim Mottagui-Tabar and Leif A Isaksson. *EMBO J.* 15, 1696–1704.
The efficiency of translation termination at NNN NNN UGA A stop codon contexts has been determined in *Escherichia coli*. No general effects are found that can be attributed directly to the mRNA sequence itself. Instead, termination is influenced primarily by the amino acids at the C-terminal end of the nascent peptide, which are specified by the two codons at the 5′ side of UGA. For the penultimate amino acid (*–2* location), charge and hydrophobicity are important. For the last amino acid (–1 location), α-helical, β-strand and reverse turn propensities are determining factors. The van der Waals volume of the last amino acid can affect the relative efficiency of stop codon readthrough by the wild-type and suppressor forms of $tRNA^{Trp}$ (CAA). The influence of the –1 and *–2* amino acids is cooperative. Accumulation of an mRNA degradation intermediate indicates mRNA protection by pausing ribosomes at contexts that give inefficient UGA termination. Highly expressed *E. coli* genes with the UGA A termination signal encode C-terminal amino acids that favour efficient termination. This restriction is not found for poorly expressed genes.
1 April 1996. *The EMBO Journal.*

☐ Principles of chaperone-assisted protein folding: differences between ***in vitro*** and ***in vivo*** mechanisms. Judith Frydman and F Ulrich Hartl. *Science* 272, 1497–1502.
Molecular chaperones in the eukaryotic cytosol were shown to interact differently with chemically denatured proteins and their newly translated counterparts. During refolding from denaturant, actin partitioned freely between 70-kilodalton heat shock protein, the bulk cytosol, and the chaperonin TCPI ring complex. In contrast, during cell-free translation, the chaperones were recruited to the elongating polypeptide and protected it from exposure to the bulk cytosol during folding. Post-translational cycling between chaperone-bound and free states was observed with subunits of oligomeric proteins and with aberrant polypeptides; this cycling allowed the subunits to assemble and the aberrant polypeptides to be degraded. Thus, folding, oligomerization and degradation are linked hierarchically to ensure the correct fate of newly synthesized polypeptides.
7 June 1996. *Science.*

☐ The kinetic folding pathway of the ***Tetrahymena*** ribozyme reveals possible similarities between RNA and protein folding. Patrick P Zarrinkar and James R Williamson. *Nat. Struct. Biol.* 3, 432–438.
The authors have probed the nature of the individual kinetic steps in the folding of the *Tetrahymena* ribozyme by studying the folding kinetics of mutant ribozymes. After rapid formation of the first structural subdomain, a slow step precedes stable formation of the second subdomain. The rapid and early formation of short-range secondary structure, the hierarchical formation of kinetic folding units corresponding to structural subdomains, and the formation of tertiary interactions between subdomains late during the folding process appear to be common features of the folding mechanism for both RNA and proteins.
May 1996. *Nature Structural Biology.*

☐ Specificity of ribonucleoprotein interaction determined by RNA folding during complex formation. Frédéric H-T Allain, Charles C Gubser, Peter WA Howe, Kiyoshi Nagai, David Neuhaus and Gabriele Varani. *Nature* 380, 646–650.

The human U1A protein binds an RNA hairpin during splicing and regulates its own expression by binding an internal loop in the 3′-untranslated region of its pre-mRNA, preventing polyadenylation. Here, the authors report the nuclear magnetic resonance structure of the complex between the regulatory element of the U1A 3′-untranslated region and the U1A protein RNA-binding domain. Specific intermolecular recognition requires the interaction of the variable loops of the ribonucleoprotein domain with the well structured helical regions of the RNA. Formation of the complex then orders the flexible RNA single-stranded loop against the protein β-sheet surface and reorganizes the carboxy-terminal region of the protein to maximize surface complementarity and functional group recognition.
18 April 1996. *Nature.*

□ Ribozyme-catalysed amino-acid transfer reactions. Peter A Lohse and Jack W Szostak. *Nature* 381, 442–444.

The authors have used *in vitro* selection and evolution to isolate ribozymes with acyl transferase activity from a pool of random RNA sequences. One of these acyl transferases with a 5′-amino group transfers an amino acid to itself in a reaction that the authors propose to be analogous to peptidyl transfer on the ribosome.
30 May 1996. *Nature.*

□ Structural basis of ligand discrimination by two related RNA aptamers resolved by NMR spectroscopy. Yinshan Yang, Michel Kochoyan, Petra Burgstaller, Eric Westhof and Michael Famulok. *Science* 272, 1343–1347.

In a previous study, an RNA aptamer for the specific recognition of arginine was evolved from a parent sequence that bound citrulline specifically. The two RNAs differ at only three positions out of 44. The solution structures of the two aptamers complexed to their cognate amino acids have now been determined by two-dimensional nuclear magnetic resonance spectroscopy. Both aptamers contain two asymmetrical internal loops that are not well ordered in the free RNA but that fold into a compact structure upon ligand binding.
31 May 1996. *Science.*